A Theoretical Model for Intraperitoneal Delivery of Cisplatin and the Effect of Hyperthermia on Drug Penetration Distance[1]

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Abstract

A theoretical model for the intraperitoneal (i.p.) delivery of cisplatin and heat to tumor metastases in tissues adjacent to the peritoneal cavity is presented. The penetration distance (the depth to which drug diffuses directly from the cavity into tissues) is predicted to be on the order of 0.5 mm. The model shows that exchange with the microvasculature has more effect than cellular uptake in limiting the penetration distance. Possible effects of hyperthermia are simulated, including increased cell uptake of drug, increased cell kill at a given level of intracellular drug, and decreased microvascular density. The model suggests that the experimental finding of elevated intracellular platinum levels up to a depth of 3 to 5 mm when drug is delivered i.p. by a heated infusion solution is due to penetration of heat to this distance, causing increased cell uptake of drug. Beyond a depth of about 0.5 mm, the drug is delivered mainly through the circulation. Use of sodium thiosulfate to deactivate systemic cisplatin may therefore be counterproductive when heat is delivered locally. The model suggests that i.p. delivery of heat, combined with systemic delivery of drug, may be as effective as i.p. delivery of heat and drug.

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Introduction

Intraperitoneal (i.p.) delivery of chemotherapeutic drugs such as cisplatin was developed for diseases such as ovarian carcinoma, which metastasize mainly in the peritoneal area. Inconsistent results have been obtained in clinical trials that compared this therapy to intravenous (i.v.) delivery. Alberts et al. [2] reported that i.p. therapy gave superior clinical results compared to i.v. therapy, but Polyzos et al. [52] were not able to reproduce this finding. Because results have been mixed, researchers have looked to regional hyperthermia as a way to improve i.p. therapy. Leopold et al. [37], using external radiation to achieve locally elevated temperatures, were able to achieve a mean temperature of only 40.7°C, which they viewed as insufficient to achieve successful enhancement of therapy. More recently, the technique of continuous hyperthermic peritoneal perfusion has been introduced to overcome this problem. Steller et al. [58] performed a pilot Phase I trial, and were able to achieve i.p. temperatures of 41°C to 43°C, whereas Bartlett et al. [3], in another Phase I trial, reported 42°C to 43°C. Clinical success in one case of malignant mesothelioma of the peritoneum was reported by de Bree et al. [8], and Park et al. [50] also found encouraging results for this disease. More recently, Takahashi et al. [60] reported that intraoperative hyperthermic peritoneal lavage was clinically beneficial for gastric cancer.

The rationale for using i.p. therapy is that it allows selective delivery of cisplatin to tissues adjacent to the peritoneal cavity. Such tissues may receive drug both from the peritoneal cavity and from the vascular system. The extent of selective delivery is conveniently expressed as penetration distance, which may be defined in several different ways. Van de Vaart et al. [62] defined penetration distance as the depth from the peritoneal cavity surface into the tissues within which drug concentration at a given time point, as measured by the number of platinum–DNA adducts, is greater than that in control cells distant from the peritoneal cavity. An alternative definition [40] is the distance within which the concentration of drug is significantly higher with i.p. therapy than would be achieved if an amount of drug resulting in the same plasma area under the concentration–time curve (AUC) was delivered intravenously. In the present study, penetration distance is defined as the distance at which the peak concentration first approaches its plateau value to within 1% deviation. Regardless of which definition of penetration distance is used, it is clear that beyond this distance, i.p. delivery is not necessarily more advantageous than i.v. delivery. Los et al. [40] sectioned rat peritoneal tissue 168 hours after i.p. and i.v. exposures and found that drug levels were elevated with i.p. delivery relative to i.v. up to a distance of only 1.5 mm from the peritoneal surface. Metastases greater than this size would presumably not receive higher drug exposure from i.p. therapy. Polyzos et al. [52] speculated that the size of metastases is a factor determining whether i.p. therapy is more effective than i.v. therapy.

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In studies combining hyperthermia with i.p. delivery, the rationale has been that use of hyperthermia enhances the penetration of drug from the i.p. space [3,5,58,62]. Apparently supporting this assumption, van der Vaart et al. [62], in a clinical trial on ovarian cancer patients, found that platinum–DNA adduct levels were elevated at a depth of 3 to 5 mm into tumor nodules, compared to control levels determined from buccal cells, which received only systemic exposure. In this case, the heat was delivered locally by infusing the peritoneal cavity with perfusate at an elevated temperature, and the elevation of adduct levels may actually reflect the penetration distance of heat, as discussed below.

The overall goal of the present study is to estimate the penetration distance of intraperitoneally delivered cisplatin into surrounding tissues, and to predict the effects of hyperthemia on the penetration distance. A previous theoretical model for drug penetration distance in i.p. therapy was presented by Dedrick and Flessner [10], who estimated the penetration distance as $(D/k)^{1/2}$, where D is drug diffusivity and k is a first-order rate constant describing blood uptake by microvessels in the tissues. This estimate neglects several potentially significant effects. Penetration may be limited by cellular uptake of drug, in addition to microvascular uptake. Because cellular uptake is nonlinear, it cannot easily be described by a first-order rate constant. With time, drug levels build up in the circulation and the concentration difference driving transport across the microvessel wall decreases. Finally, the concentration in the peritoneal cavity is not a constant as assumed in the estimate of Dedrick and Flessner [10] but decreases with time. Although some of these factors were considered subsequently in more detailed theoretical modeling of Flessner et al. [19], their model was developed primarily for sucrose, and therefore did not incorporate particular properties of cisplatin such as cell uptake kinetics and peritoneal cavity half-life. These factors are considered in the model presented below, which is developed specifically for cisplatin.

Several mechanisms can be proposed by which hyperthermia might alter the penetration distance of cisplatin from the peritoneal cavity into tissues. One possibility is that hyperthermia increases transport parameters such as drug diffusivity in tissues or hydraulic conductivity of tissues. Alternatively, hyperthermia may change the cellular uptake of drugs; however, because studies have shown that uptake is increased with heat, this would actually lead to decreased penetration. A further possibility is that at high temperatures, tissue perfusion or microvascular surface density changes. A decrease in microvascular surface density could lead to less uptake of drugs by microvessels, which would tend to increase the penetration distance, whereas an increase would have the opposite effect. Hyperthermia also appears to cause higher cytotoxicity for the same amount of extracellular drug exposure. Although this effect could lead to cell kill extending a greater depth into tissues, this is not due to increased penetration of cisplatin, but to the penetration of heat.

In the present study, a theoretical model for i.p. delivery of cisplatin in rats is developed. The model is used to predict the penetration distance of the drug from the peritoneal cavity into the tissues. The penetration distance of hyperthermia delivered by infusing heated solutions into the peritoneal cavity is estimated. Possible effects of hyperthermia on drug transport, including increased cellular uptake and cytotoxicity and reduced microvascular density, are considered. Also, the effect of deactivating cisplatin in plasma using sodium thiosulfate is simulated. This approach has been used experimentally to reduce systemic exposure and associated host toxicity [3,11,23].

Materials and Methods

Cellular Pharmacodynamics

A mathematical model of mass transport near the peritoneum was developed to predict the concentration of cisplatin in the tissues adjoining the peritoneum as a function of time. Parameter values were chosen for the rat, based on availability of pharmacokinetic data for this species. The applicability of the model for humans was also considered. The penetration of the drug into tissues depends on the rate of cellular uptake, which is assumed to be described by the cellular pharmacokinetic model of El Kareh and Secomb [13]:

$$\frac{dc_1}{dt} = k_1 c_e - k_1' c_i \tag{1}$$

$$\frac{dc_k}{dt} = k_2 c_1 - k_3 c_k \tag{2}$$

where c_e is the extracellular concentration of free platinum, and c_i and c_k are the intracellular concentrations of total platinum and DNA-bound platinum (the species responsible for cell kill), respectively. Experimental methods measure intracellular platinum levels rather than cisplatin levels, and cisplatin forms other active platinum species. All concentration variables in the model are assumed to refer to ultrafilterable platinum (i.e., platinum either in the form of cisplatin or its low-molecular-weight metabolites). Values for the parameters k_1, k_1', k_2, and k_3 were derived [13] by fitting experimental data [38] for ovarian cancer cells. Values and sources of all model parameters are given in Table 1. Cytotoxicity is described by the following pharmacodynamic model [13]:

$$S = \frac{1}{1 + A(c_k^{\text{peak}})^m} \tag{3}$$

where S is survival fraction, and c_k^{peak} is the peak value (over time) of c_k. The parameter A cannot be estimated because data on bound intracellular concentration are not available. A value $A = 1$ was therefore assigned [13], and the absolute levels of c_k are undetermined. Predicted values express relative changes only.

Simulation of Drug Transport into Tissues

Because drug penetration distance is small [40], the peritoneal surface can be approximated by a plane. The transport of cisplatin into the tissues is then governed by:

$$\frac{\partial c_e}{\partial t} = D\frac{\partial^2 c_e}{\partial x^2} - \frac{1-\phi}{\phi}(k_1 c_e - k_1' c_i) - \frac{PS_v}{\phi}(c_e - c_b) \quad (4)$$

where x represents distance into the tissues starting at the peritoneal cavity surface, t is time, D is the diffusivity of cisplatin in tissues, and ϕ is the volume fraction of extracellular space in the tissues. The factors $(1-\phi)/\phi$ and $1/\phi$ are included to account for the fact that intracellular concentration is expressed as micrograms Pt per milliliter of intracellular space, and extracellular concentration as micrograms Pt per milliliter of extracellular space.

The concentration in the peritoneal cavity as a function of time, $c_{ip}(t)$, was obtained from Figure 4 of Los et al. [40], with linear interpolation between the given data points. These data were used to predict the concentration of platinum in the plasma, $c_b(t)$, using the following equation:

$$\frac{dc_b}{dt} = \frac{PS_V}{V_d}A_P\int_0^{\delta}(c_e - c_b)dx - k_b c_b + \frac{F_{lym}}{V_d}c_{ip}(t) \quad (5)$$

where P is the microvascular permeability, S_v is the surface area of perfused microvessels per unit volume of the tissue surrounding the peritoneum, V_d is the volume of distribution of cisplatin in plasma, and F_{lym} is the rate of lymph flow directly from the cavity into the plasma. The integral represents uptake by the microcirculation from the region that is supplied directly from the peritoneal cavity. The distance δ is defined by the point at which the spatial derivative of c_e is less than a small fraction of its maximum value. This ensures that the region of integration includes all tissues that are significantly affected by drug transport from the peritoneal space. The constant k_b describes the rate of platinum clearance from the blood by tissues throughout the body. The predictions of this model were compared with experimental data [40].

Values of V_d and k_b were obtained from a study of pharmacokinetics of cisplatin in humans with i.v. administration [51]. The value of the volume of distribution, which depends both on total blood volume and on the extent of binding of the drug to plasma proteins or other plasma components, was assumed to scale primarily with body weight (Table 1). Based on the findings of Dedrick [9] that the half-life of methotrexate showed only weak dependence on body weight (slope 0.2), the value of k_b of Patton et al. [51] was assumed to apply also to the rat. Values of S_v, P, and ϕ that were measured in some (nontumor) tissues lining the peritoneum were used. Because the peritoneum consists of several different tissue types, it is possible that these parameters vary from one location to another—an effect that is

Table 1. Model Parameters.

Symbol	Description	Value	Source
MW_{cis}	Molecular weight of cisplatin	300	–
MW_{Pt}	Molecular weight of platinum	195	–
D	Diffusion of cisplatin in tissues	2.5×10^{-6} cm^2/sec	Correlation of [59] (normal tissues)
φ	Extracellular space volume fraction in tissue	0.3	Representative value based on values for various peritoneal tissues from Ref. [21]
W_{rat}	Weight of rat	200 g	[35]
W_{human}	Weight of human	70 kg	–
V_d	Volume of distribution of cisplatin in plasma	57,500 cm^2 (human), (0.2/70) $\times$ 57,500 cm^2 (rat)	Ref. [51] for human; rat value assumed to be scaled by weight
$T_{1/2}^{ip}$	Half-life of cisplatin in peritoneal cavity	1.8 hours (human); 1 hour (rat)	[40,47] (rat)
k_b	Rate constant for plasma drug clearance	0.0248 min^{-1}	[51] (see text)
A_P	Wetted peritoneal surface area	5500 m^2 (human); 200 cm^2 (rat)	Ref. [6] for human; Ref. [35] for rat
P	Microvascular permeability of cisplatin, or other ultrafilterable Pt species	1.43×10^{-4} cm/sec	Ref. [28] for sucrose (MW = 342) in frog mesentery
S_v	Microvascular surface area per unit volume	70 cm^{-1}	[18]
k	Tissue thermal conductivity	2.2×10^4 g cm/(sec^3 °C)	[30]
ρ	Density of tissue or plasma	1 g/ml	Approximated by density of water
c_p	Heat capacity of tissue or plasma	4.2×10^7 cm^2/(sec^2 °C)	[7]
w_b	Blood perfusion rate	0.001667 hr^{-1}	Ref. [15] from intermediate value between 15 and 18 ml/min/100g (tumor periphery) and 2–4 ml/min/100g (tumor interior)
T_a	Arterial temperature during hyperthermia	37° C	Assumed equal to normal body temperature
F_{lym}	Lymph flow rate	0.16 ml/hr	[20]
k_1	Cisplatin cellular pharmacodynamic model parameter (for ovarian cancer cells)	0.327 hr^{-1} at 37°C; 0.529 hr^{-1} at 43°C	Ref. [13] fit to data of Ref. [38]; Ref. [34] (see text)
k_i	Cisplatin cellular pharmacodynamic model parameter (for ovarian cancer cells)	0.02413 hr^{-1}	Ref. [13] fit to data of Ref. [38]
k_2	Cisplatin cellular pharmacodynamic model parameter (for ovarian cancer cells)	3.745 hr^{-1} at 37°C; 28.07 hr^{-1} at 43°C	Ref. [13] fit to data of [38]; Ref. [41] (see text)
k_3	Cisplatin cellular pharmacodynamic model parameter (for ovarian cancer cells)	1.441 hr^{-1}	Ref. [13] fit to data of Ref. [38]
m	Cisplatin cellular pharmacodynamic model parameter (for ovarian cancer cells)	1.956	Ref. [13] fit to data of Ref. [38]
A	Cisplatin cellular pharmacodynamic model parameter (for ovarian cancer cells)	1 (μg/ml)$^{-1}$	Ref. [13] fit to data of Ref. [38]

neglected here. A value for D specific to peritoneal tissues could not be found, and therefore a general correlation with molecular weight for tissues was used (Table 1). The possibility that S_v, P, D, and ϕ may vary between tumor and normal tissues [29] is also neglected here.

The case in which sodium thiosulfate is administered intravenously to reduce systemic exposure is also simulated. In this case, the plasma concentration of platinum (the platinum that is present in active platinum species) is assumed to be zero at all times. In reality, only part of the systemic platinum may be deactivated, so the model results may overestimate the effect of sodium thiosulfate administration.

To determine the significance of convective transport of drug relative to diffusive transport, the dimensionless groups $u(T/D)^{1/2}$ and $u(Dk)^{-1/2}$ were estimated, where u is a typical fluid velocity out of the cavity, D is the diffusivity of drug in the tissues, T is the half-life of drug in the cavity, and k is the rate of uptake by microvessels, cells, protein binding, etc., per unit volume of tissue. Both these dimensionless groups represent the ratio of the penetration distance for drug resulting from convection to that resulting from diffusion. The first group is for the case in which penetration is limited by the half-life of the drug in the cavity; the second is for the case in which penetration is limited by uptake of the drug in the tissues. It is not clear *a priori* which of these two factors is dominant in limiting penetration. The convective velocity u is estimated as follows. Flessner and Schwab [22] obtained a fluid loss rate of 5 ml/hr from the rat peritoneal cavity after infusion of an isotonic solution. The rat peritoneal surface area is approximately 450 cm^2 (Table 1), which implies a flow velocity of $u = 3.1 \times 10^{-6}$ cm/sec. Using the half-life $T = 1.8$ hours [47] (Table 1) gives $u(T/D)^{1/2} = 0.06$. If k is approximated as the rate of uptake due to microvessels, it can be estimated as $k = PS_v = 4.4 \times 10^{-4}$ sec^{-1}, so that $u(Dk)^{-1/2} = 0.03$. Based on these arguments, convection of drug was assumed to be negligible relative to diffusion.

The partial differential equation (Eq. (4)) was solved simultaneously with the ordinary differential equations (Eqs. 1, 2, and 5) by discretizing the spatial coordinate x on the interval $0 < x < L$, using central differences. The Crank–Nicholson method was used to discretize in time. The resulting system of coupled linear algebraic equations was solved using the LINPACK package for each time point. The distance L was taken to be large enough so that plateau values were unaffected by changing L, except in calculations where uptake by the microvasculature was set to zero. In that case, no plateau was reached at large x, and 10 cm was taken as sufficiently large. The nodes were spaced more closely near the cavity surface, the spacing starting at 0.005 cm and increasing with distance from the cavity. Time steps of 0.005 hour were determined to be sufficiently small so that halving the step size did not change the results significantly. Initial conditions were that all concentration variables were 0 at time 0, and the boundary conditions were $c_e = c_{ip}(t)$ at $x = 0$ (the cavity surface), and $dc_e / dx = 0$ at $x = L$. To determine the peak values of the concentration variables, simulations of 35 hours (or shorter, in the cases where equilibrium spatial distributions were achieved) were found to be adequate. For each case considered, the penetration distance d was estimated as the distance at which the peak concentration approached its plateau value within 1%.

Simulation of Effects of Hyperthermia

In the earliest studies combining hyperthermia with i.p. drug infusion, heat was administered from extracorporeal sources [37], but more recent studies have focused on delivering heat by infusing the peritoneum with a heated solution [3,50,58]. Both cases are considered here. In the case of "regional heating," the entire tissue region under consideration is assumed to be heated to a uniform temperature of 43°C for a duration t_d, after which it immediately cools to 37°C. Effects of transient adjustments in temperature are neglected. In the case of "i.p. heating," heat is assumed to be delivered from the fluid inside the cavity, which is maintained at either 43°C or 42°C for the duration t_d, after which it abruptly drops to 37°C. For this case, the distribution of temperature with distance from the cavity wall into the tissues must be determined. The bioheat equation [7] is used here:

$$\frac{\partial T}{\partial t} = \frac{k}{\rho c_p}\frac{\partial^2 T}{\partial x^2} + w_b(T_a - T) \qquad (6)$$

with parameters defined in Table 1. The boundary conditions are that $T = 43$°C at the peritoneal cavity surface for the heating duration t_d, returning to 37°C after this time; and $T \rightarrow 37$°C (normal body temperature) as $x \rightarrow 00$. The initial condition is that $T = 37$°C for all x. Eq. (6) with these conditions was solved analytically. A heating duration t_d of 90 minutes (as in the study of van de Vaart et al. [62]) was used. Parameter values appropriate for the rat were obtained from the literature (Table 1).

The effects of hyperthermia on tumor blood flow, which may include changes in microvascular surface density or perfusion, are complicated, showing both time dependence [31,33,45,46] and spatial dependence [1]. Both increases [44,54,56,57,63] and decreases [14,27,32] in tumor blood flow have been observed, with some studies reporting damage to supplying vessels [43]. As summarized by Reinhold and Endrich [53], most experimental tumors show a decrease or cessation of blood flow with hyperthermia, but the evidence is less conclusive for human tumors. Changes in blood flow are represented in the model by changes in the perfusion (w_b) and in the surface density of perfused vessels (S_v). Increases in these parameters would lead to decreases in the penetration distances of heat and drug, respectively, when delivered locally to the i.p. space. To examine the possibility of increased penetration distance with hyperthermia, we focus here on cases in which blood flow is decreased. The cases in which S_v is one tenth the value given in Table 1 and in which $S_v = 0$ were therefore included in Eq. (4) to model possible effects of locally reduced blood supply in the tumor. In each case, the perfusion parameter w_b was reduced by the same proportion from its value in Table 1. This assumption of proportional change of perfusion and

microvascular density was based on observations that hyperthermia causes reduced blood flow through vessel damage [14,16,43].

Changes in blood flow in normal peritoneal tissues are neglected in the present model. In reality, normal tissue blood flow may increase with heating. Effects of such behavior are considered in the Discussion section. It can be assumed that most of the peritoneal cavity is lined by normal tissues, which is therefore the source of most drug uptake by the circulation. Therefore, the plasma concentration $c_b(t)$ is estimated in all cases by simultaneous solution of (Eqs. 1, 2, 4, and 5) with $S_v = 70\ cm^{-1}$, corresponding to normal tissues. To calculate concentration profiles in the tumor, Eq. (4) is again used, with S_v values of 70 cm^{-1}, 7 cm^{-1}, and zero. This procedure neglects any gradients in the transverse direction (parallel to the cavity surface) due to gradients in blood flow and other properties between normal and tumor tissues.

Hyperthermia has been found to increase drug uptake [12,34], which tends to decrease the drug penetration distance. Kusumoto et al. [34] observed a 1.57-fold increase in intracellular platinum in HeLa cells following a 30-minute exposure to 33.2 μM cisplatin at 42.8°C, which, by linear extrapolation, gives a 1.62-fold increase at 43°C. This was modeled here by assuming that the uptake parameter k_1 increases by a factor of 1.62 at 43°C, so that the total intracellular concentration is increased by the same factor. For temperatures between 37°C and 43°C, linear interpolation using the values k_1 = 0.327 (at 37°C) and k_1 = 0.529 (at 43°C) (Table 1) was used. The factor k_1 is related to the cellular permeability. Because it is believed that heat increases cell uptake by increasing membrane permeability [48], the change in uptake at higher temperatures is logically represented by increasing the value of k_1.

Several studies have shown that heat not only causes greater cellular uptake of drugs, but also higher cytotoxicity at the same level of intracellular drug [24,34]. Using colon carcinoma cells, Los et al. [41] measured cytotoxicity *versus* cisplatin concentration, for a 1-hour drug exposure, with and without hyperthermia. At 5 μg/ml cisplatin, survival fraction decreased by a factor 130 from 0.13 at 37°C to 0.001. The effect of temperature on cytotoxicity was represented here by assuming that the parameter k_2, representing the rate of DNA binding, increases with temperature, so as to decrease survival fraction by a factor of 100 with heating at an extracellular concentration of 5 μg/ml cisplatin and 1-hour exposure. In reality, the effects of heating may result from changes in both DNA adduct formation and adduct processing [25]. Here, the changes in k_2 are assumed to represent the combined effects of both processes. Under these assumptions, k_2 was estimated to increase from 3.745 hr^{-1} at 37°C to 28.1 hr^{-1} at 43°C. Linear interpolation between these two values was used to determine k_2 as a function of temperature between 37°C and 43°C. For comparison, the increase in k_2 with heat could also be estimated in a similar manner from the data of Hettinga et al. [25] for murine Ehrlich ascites cells, giving 25.5 hr^{-1} at 43°C, close to the estimate of 28.1 hr^{-1} obtained from the Los et al. data. The latter value was used in simulations here.

Several other simplifying assumptions were made. In simulations involving time-dependent temperature changes, the values of k_1 and k_2 were assumed to depend only on the instantaneous temperature, and any time-delayed effects of temperature changes were not included. Protein binding of cisplatin in the tissues was neglected. The peritoneum itself was considered to be identical to the adjacent tissues in terms of transport properties. Some researchers have assigned a special resistance to this membrane [55], but Flessner [17] concluded that "the peritoneum is similar to the underlying tissue with respect to its transport resistance to small solutes." Also, it was assumed that the cellular uptake of all tissues adjacent to the cavity could be modeled using the cellular pharmacokinetics of ovarian carcinoma cells. In actuality, some of the cells in this region will be normal, and therefore may have different cellular drug uptake. Finally, the lymph flow coming directly from the cavity is assumed to instantly enter the circulation, whereas in reality, there is a time delay. However, the lymph flow term was found to have only a very small effect on the plasma concentration.

Results

Drug Penetration and Cell Survival without Hyperthermia

Figure 1 shows experimental values of plasma concentration of platinum for i.p. infusion from Ref. [40], together with the values predicted by the present model. The agreement is satisfactory. Further simulations were performed for values of microvascular surface density other than S_v = 70 cm^{-1}, and predicted values then agreed less well with the experimental values.

Figure 2 shows the predicted variation of peak extracellular and bound intracellular drug levels, and of cell survival fraction with distance into the tissues. The peak value is reached at different times at different locations, so these curves do not represent profiles at any single instant in time. Except for the case of no perfusion, both concentrations approach plateau values with increasing distance, representing supply from the systemic circulation. With normal

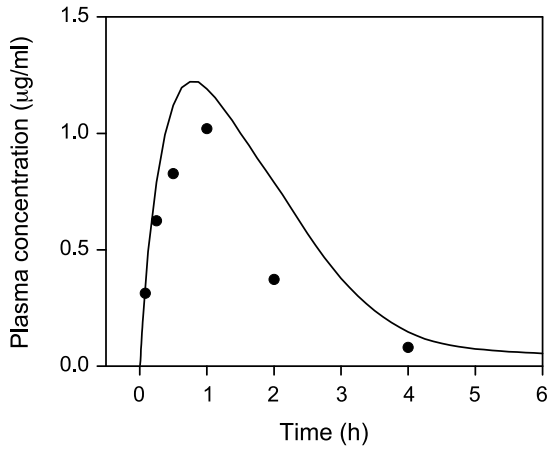

Figure 1. *Plasma concentration of (ultrafilterable) platinum after i.p. infusion. Line: theoretical model for ovarian carcinoma cells. Filled circles: experimental data for rat from Ref. [40].*

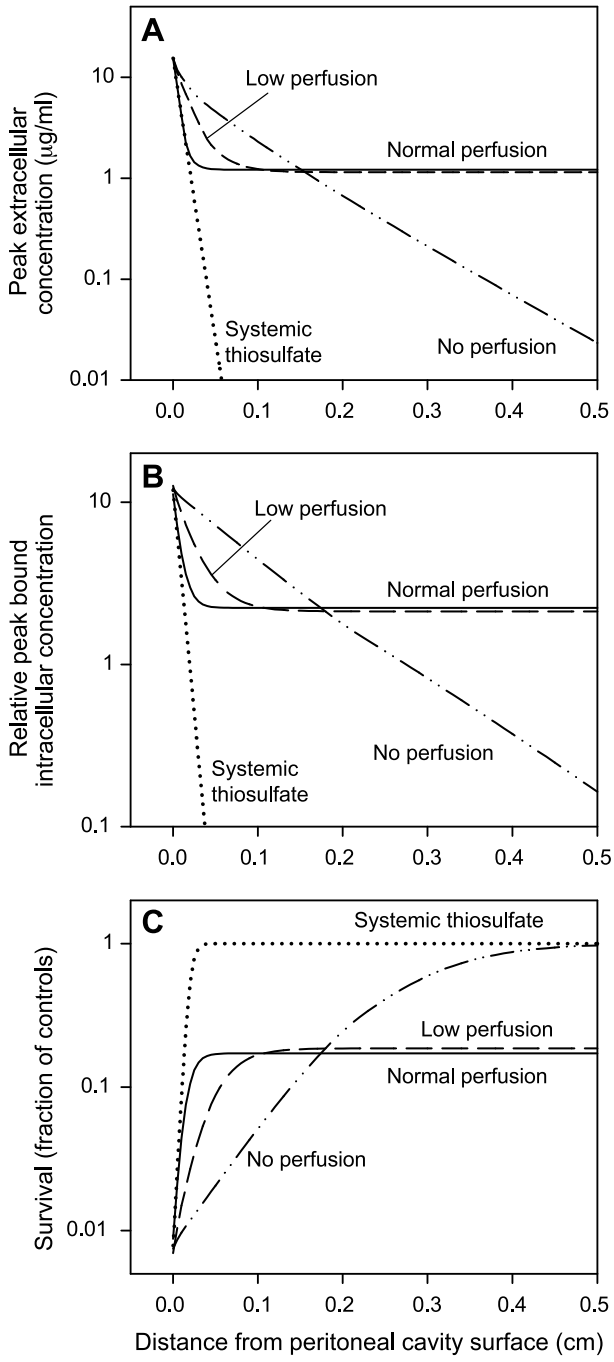

Figure 2. *Predictions of the mathematical model for rat, with ovarian carcinoma cells, without hyperthermia. Solid lines: normal tumor perfusion (vessel surface density S_v = 70 cm^{-1}). Dashed lines: low tumor perfusion, represented by reduced surface density (S_v = 7 cm^{-1}). Dashed–dotted lines: zero tumor perfusion. Dotted lines: with sodium thiosulfate administered intravenously, assumed to deactivate all circulating cisplatin. (A) Peak extracellular concentration in tumor as a function of position. The peak value is reached at different times at different points, so this curve is not a concentration profile at one instant in time. (B) Peak intracellular concentration in tumor as a function of position. (C) Survival fraction of cancer cells.*

perfusion, the penetration depth as defined above is 0.48 mm. The penetration distance based on extracellular concentration is the same. Beyond this distance, the tissue receives drug essentially only from the circulation. Although normal perfusion (w_b = 0.001667 hr^{-1}, S_v = 70 cm^{-1}) is expected in the absence of heating, results for low perfused vascular density (w_b = 0.0001667 hr^{-1} , S_v = 7 cm^{-1}) and no perfusion ($w_b = S_v = 0$) are included for comparison. With reduced perfusion, the penetration depth increases to 1.5 mm. Conversely, increasing tumor blood flow causes decreased penetration distance (results not shown). In the absence of perfusion, a penetration distance for extracellular drug as defined earlier cannot be estimated because no plateau level of concentration is reached. In that case, the peak extracellular level of drug is higher than that achieved by systemic delivery (normal perfusion curve) for a depth of up to about 1.5 mm, but is lower beyond this distance (Figure 2*A*). Corresponding behavior is seen for the peak bound intracellular concentration (Figure 2*B*) and the survival fraction (Figure 2*C*). Figure 2 also shows the effect of systemic sodium thiosulfate administration, which has been proposed as a method to decrease systemic toxicity of the therapy by deactivating the drug in the circulation. In this case, drug levels rapidly decrease to zero with distance into tissues because systemic delivery is suppressed.

Drug Penetration and Cell Survival with Hyperthermia

Figure 3 shows results corresponding to those in Figure 2 for the case of regional heating, in which all tissues adjacent to the cavity are heated to 43°C for the duration of hyperthermia, t_d = 90 minutes [3], after which they return to 37°C. Results for normal perfusion without heat (Figure 2) are included for comparison. The distribution of peak extracellular concentration (Figure 3*A*) is virtually unaffected by heating, showing that the increased cellular uptake does not significantly decrease the penetration distance. Microvascular uptake is therefore the major factor limiting drug penetration, and the penetration depth for extracellular drug is 0.48 mm, as before. Effects of reduced or zero perfusion are similar to those seen in the absence of heating. The increased levels of peak bound intracellular concentration, relative to those with no heat (Figure 3*B*), reflect the effects of both enhanced cellular uptake and enhanced intracellular binding (increased k_1 and k_2). Predicted survival fractions (Figure 3*C*) are reduced by about two orders of magnitude in the presence of regional heating.

Figure 4 shows the temperature profile in peritoneal tissues, assuming that heat is delivered by infusing the peritoneal cavity (i.p. heating) with a solution at temperature 43°C or 42°C [62]. With normal values (at 37°C) of perfusion w_b in the tissues, the profile reaches equilibrium at around 5 minutes. Significant enhancement of cell kill occurs at temperatures above about 40°C to 41°C [61], which occur here up to depths of about 0.5 cm with normal perfusion. With reduced perfusion (w_b one tenth of normal), an equilibrium profile is reached after about 8 minutes, and the penetration distance of heat is increased. With no perfusion (w_b = 0), no equilibrium is reached and the depth of penetration of heat increases indefinitely with time. Results are shown for 90 minutes of heating. For reduced and zero perfusion, therapeutically useful temperatures (above 40°C) are achieved up to depths of 1 to 2 cm.

In Figure 5, model predictions corresponding to those in Figure 3 are shown for i.p. heating. Cellular uptake and

intracellular binding are functions of temperature as described previously, and results are shown for normal, reduced, and zero perfusion. Results for normal perfusion without heat (Figure 2) are again included for comparison. These results show increased intracellular binding (Figure 5*B*) and cytotoxicity (Figure 5*C*) near the peritoneal surface, resulting from the combined effects of the temperature-dependent changes in pharmacodynamic and transport parameters. At depths of 2 cm or more, concentrations closely approach the plateau level, and this distance represents an upper limit on the predicted extent of therapeutic benefit from i.p. heating.

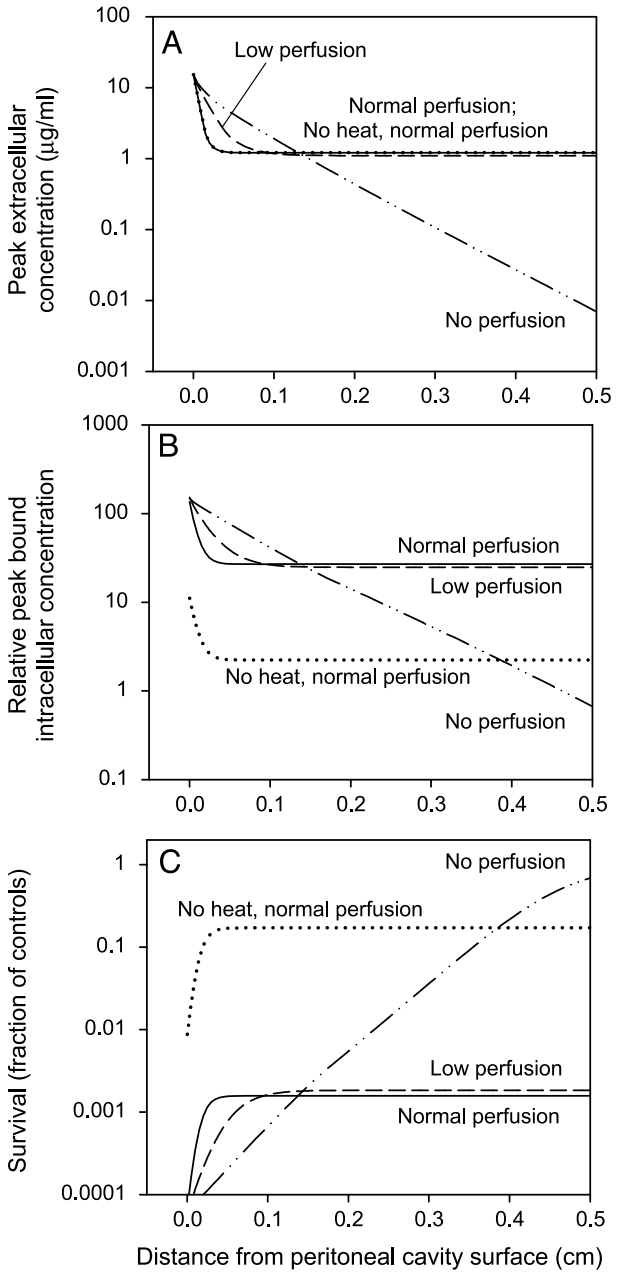

Figure 3. *Predictions of mathematical model as in Figure 2, but with regional hyperthermia (i.e., all tissues adjacent to the peritoneal cavity heated to a uniform 43 C). Solid lines: normal tumor perfusion. Dashed lines: low tumor perfusion. Dashed–dotted lines: zero tumor perfusion. Dotted lines: normal tumor perfusion without heating, as shown in Figure 2.*

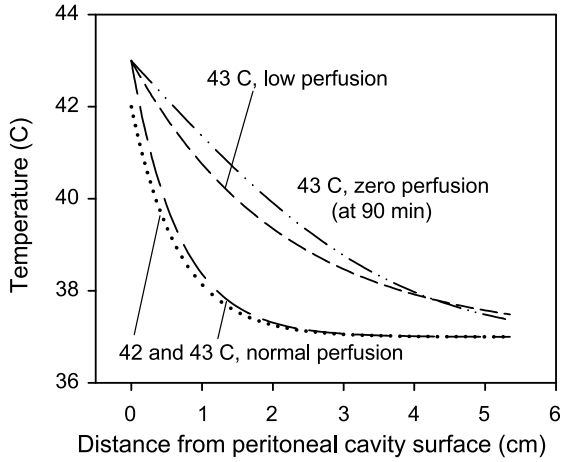

Figure 4. *Predictions of theoretical model for penetration of heat into the tumor tissues adjacent to the peritoneal cavity. The cavity is assumed to be at a constant temperature of either 42°C or 43°C for 90 minutes, after which it returns to 37°C. The cases of normal, low, and zero tumor perfusion are shown. Except for the case of $w_b = 0$, all profiles are steady state reached after approximately 5 to 10 minutes.*

Discussion

In the present model, the accumulation of drug in the plasma following i.p. infusion is assumed to result from uptake by microvessels in tissues adjacent to the peritoneal cavity. According to Eq. (5), the predicted plasma concentration depends critically on the parameters describing transport and exchange of the drug in the tissues. The agreement between predicted and observed plasma concentrations of ultrafilterable platinum (Figure 1) therefore supports the validity of the assumed model for tissue transport, which is used to estimate the penetration distance. The results of Figure 1 were for an assumed value of microvascular surface density of $S_v = 70\ \text{cm}^{-1}$ (Table 1), and agreement is considerably poorer if the value of S_v is changed substantially. This suggests that this value of S_v is representative, on average, of the tissues lining the peritoneal cavity. However, the cavity is lined with a number of different tissues, and local deviations from this average value may occur.

The model predicts that the penetration distance for drug diffusing directly from the cavity into the surrounding tissues is small, on the order of 0.48 mm, for normally perfused tissue (Figure 2). The basic reason for this result is that, in perfused tissues, the microvasculature provides a large area for exchange, and extracellular concentration therefore tends to be equilibrated with plasma concentration. Drug diffusing into the tissues at higher concentrations is rapidly taken up by the microvessels. The plateau levels in Figure 2 at depths beyond roughly 0.48 mm reflect systemic levels. The estimate of penetration depth given by the formula $(D/k)^{1/2}$ [10], with $k = PS_v$ and parameter values from Table 1, is 0.14 mm, comparable to the predictions of the present model. The theoretical predictions of Flessner et al. [19] for tissue concentrations of intraperitoneally administered sucrose show a comparable penetration distance, as do the experimental and theoretical concentration profiles for EDTA of Flessner et al. [21], which level off at about 0.3 to

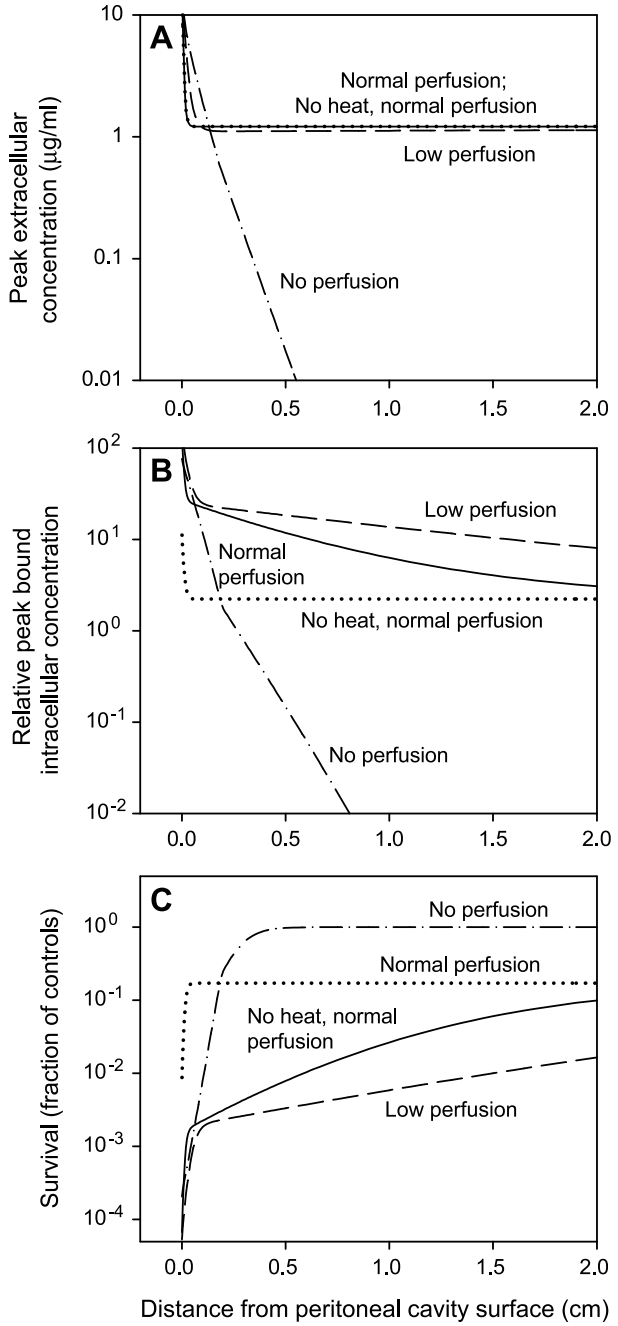

Figure 5. *Predictions of mathematical model as in Figure 2, but with i.p. hyperthermia, achieved by infusing the peritoneal cavity with a solution at 43°C. Solid lines: normal tumor perfusion. Dashed lines: low tumor perfusion. Dashed–dotted lines: zero tumor perfusion. Dotted lines: normal tumor perfusion without heating, as shown in Figure 2.*

0.4 mm from the peritoneal surface in uterine, liver, and spleen tissues, and at about 0.5 to 0.6 mm in parietal tissues.

Experimental studies have yielded widely varying estimates of the penetration depth. Ozols et al. [49] compared i.p. with i.v. delivery of adriamycin in mice. Intense fluorescence associated with levels of adriamycin higher than that achieved by i.v. exposure could be seen only in the outermost four to six cell layers of intra-abdominal tumors. This implies a penetration distance around 0.1 mm, based on a cell dimension of no more than 20 μm. In a study comparing i.p. to i.v. delivery of cisplatin in rats, Los et al. [40] measured levels of platinum in i.p. tumors at 4, 24, and 168 hours after treatment, for both single cycles and fractionated doses. No difference was found at 4 and 24 hours. At 168 hours, with a fractionated dose, the amount of platinum per weight of tumor tissue was found to be significantly higher for i.p. than that for i.v. therapy up to a distance of 1.5 mm into the tumor. However, by 24 hours after i.p. therapy, plasma concentrations of platinum were higher than concentrations in the peritoneal cavity, so it is not clear that the apparent regional advantage at 168 hours could have resulted from diffusion directly into tissues from the i.p. space. Los et al. [42] analyzed the spatial distribution of platinum 24 hours after the last injection of drug, and found that levels at 1 mm were substantially elevated over levels at 3 mm. Los and McVie [39] summarize other studies as giving estimates for the penetration depth of cisplatin in the 1 to 3 mm, or 2 to 2.5 mm range. One possible explanation for the observation of penetration depths larger than predicted here is that microvascular surface density levels in the vicinity of these nodules may be lower than assumed (S_v = 70 cm^{-1}). However, as already mentioned, the assumed value is consistent with drug concentrations observed in plasma. Another possibility is that spatial gradients in the volume fraction φ of extracellular space [21] significantly affect the concentration distribution. Here, φ is held constant and this possible effect is excluded.

The possible effects of hyperthermia on drug distribution and cytotoxicity in peritoneal tissues are explored in Figures 2, 3, and 5. As discussed above, some studies have indicated decreased tumor blood flow, or even vascular stasis, in tumors in rodent models with high heat [53,63]. Figure 2 shows that eliminating microvascular uptake, which may be at least partially achievable by hyperthermia or by an agent that targets tumor vasculature, such as combrestatin [26], would lead to reduced tumor cell survival up to a depth of about 1.7 mm, but to increased survival beyond this depth. If high levels of cisplatin could be maintained for a longer time in the cavity in the absence of microvascular uptake, the drug would be able to diffuse out further. However, this might result in unacceptable host toxicity. Hyperthermia has also been shown to result in increased cell uptake [12]. Although this might be expected to result in a decreased penetration distance, the model results (Figure 3) indicate no significant change in penetration distance.

Several authors [5,41,62] have stated that the penetration depth of cisplatin is increased under hyperthermic conditions. Possible mechanisms by which hyperthermia could affect drug penetration are effects on convection and diffusion in tissues, cell uptake of drug, tumor blood flow, and vascular permeability to drug. Because diffusion is dominant over convection, changes in convection, due for example to tissue expansion or degradation of the extracellular matrix, are unlikely to make a significant difference. Large changes in tissue diffusivity with heating from 37°C to 43°C are also unlikely. Figure 3 shows that changes in cell uptake with heat have negligible effect on penetration distance of cisplatin,

although the effect might be significant for other drugs that are taken up more rapidly by cells. The remaining two possibilities (changes in microvascular density or in microvascular permeability) are mathematically equivalent because they appear in the model only in the factor PS_v in Eq. (6). Figure 3 shows that a decrease in this parameter can increase the penetration distance of the drug, at the expense of a reduction in drug levels beyond the penetration distance. However, tumor microvascular permeability has been observed to increase with hyperthermia above 43°C [36], whereas evidence for decreased perfusion in hyperthermia is not conclusive [53,57,63].

For the case of i.p. heating, the present results (Figure 5) suggest an alternative explanation for the observation of increased penetration depth with hyperthermia. Penetration of heat into the tissues (Figure 4) results in a region of increased drug uptake that extends 1 to 2 cm into the tissues. This phenomenon may underlie the observation [62] of increased levels of platinum–DNA adducts at depths greater than 3 mm in human patients, relative to control cells distant from the peritoneum, when i.p. cisplatin was accompanied by i.p. hyperthermia. Thus, the apparent increase in penetration distance may reflect penetration of heat, leading to enhanced drug uptake, without any increase in extracellular drug levels (Figure 5*A*).

Sodium thiosulfate, which deactivates cisplatin, has been administered intravenously [3,11,23] concurrently with i.p. therapy (with and without hyperthermia) on the grounds that it reduces systemic exposure. As shown in Figure 2, this treatment would have the effect of greatly reducing drug levels in tissues at points beyond the penetration distance of about 0.48 mm, where the tissues receive the drug only from the circulation. Despite studies showing that it has the potential to reduce host toxicities, sodium thiosulfate has not been generally adopted for clinical use because of concerns that it also reduces anticancer effects [4]. The present results suggest that simultaneous administration of thiosulfate with cisplatin would not be advantageous.

The results presented here are based on parameter values for the rat, some of which may differ from those for humans. The microvascular density, cellular uptake, and diffusivity of cisplatin in the tissues adjacent to the cavity can be assumed to be of similar magnitude for both species. The peritoneal half-life of cisplatin for humans [47] is similar to that for rats [40]. Penetration distances for the drug should therefore be comparable in both species. The ratio of human to rat blood volume is approximately equal to the weight ratio, 70 kg / 0.2 kg = 350, whereas the ratio of the human peritoneal surface area to that of the rat is 5500 cm^2 / 200 cm^2 = 28 (Table 1). If penetration distances are assumed equal, the total uptake of cisplatin is therefore 28 times larger for humans, assuming the same i.p. concentration. Because the blood volume is 350 times larger, plasma concentrations are expected to be lower, leading to lower plateau concentrations and higher plateau survival fractions (Figure 2). This assumes that the ratio of volume of distribution to plasma volume is approximately the same for the rat and humans.

Several simplifying assumptions were made in the model presented here. A kinetic model of cellular cisplatin uptake developed for ovarian tumor cells was used not only within the tumor but also for all normal tissues surrounding the peritoneum. However, cellular drug uptake was found to have only a slight effect on concentration profiles, so this assumption does not lead to significant errors. Another model assumption is that blood flow in the normal peritoneal tissues remains unchanged with heat. A heat-induced increase in blood flow in normal tissues during the period of i.p. drug exposure might result in increased plasma drug levels, if associated with an increase in perfused vessel density (S_v). Such an effect would increase the plateau values of concentration (Figures 2, 3, and 5), but would not affect the penetration distance. In practice, if heating resulted in increased plasma levels, a reduction in the applied dose would probably be needed to avoid increased systemic toxicity, so using the same plasma concentration function for the heated and nonheated cases would be appropriate.

In summary, the model predicts that the distance that cisplatin diffuses into peritoneal tissues is short, about 0.48 mm, and that this distance is limited by the rate of microvascular uptake. Beyond this distance, the drug reaches tissues primarily through the circulation. The model suggests that hyperthermia administered locally, in the form of a heated drug solution infused into the peritoneal cavity, does not cause increased penetration of the drug into the tissues. However, cell kill may be locally increased up to a distance on the order of 1 cm into the tissues, due to penetration of heat and the resulting increases in cellular uptake and toxicity of drug. It may therefore be therapeutically advantageous to combine i.v. drug administration with delivery of hyperthermia by i.p. infusion.

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